

The following information was provided to Washoe County from the State of Nevada Water Quality Planning and has been reviewed by the State of Nevada Department of Health and Human Services, Division of Public and Behavioral Health, Office of Public Informatics and Epidemiology.

Potentially Toxicogenic (PTOX) Cyanobacteria Report Project: Nevada State Health Lab  
Samples Received: August 22, 2018 Report Prepared: August 22, 2018 Analyst: Amanda Foss  
Sample ID Site Collected 1 Lemmon Lake Inlet 8/21/18 2 Lemmon Lake Inlet 8/21/18

Method One mL aliquots of live sample were prepared using Sedgewick Rafter cells. The samples were scanned at 100X for the presence of potentially toxigenic (PTOX) cyanobacteria using a Nikon Eclipse TE200 inverted microscope equipped with phase contrast optics. Higher magnification was used as necessary for identification and micrographs.

#### Results

1 The sample was dominated by filamentous and flagellated algae, including filamentous green algae (Chlorophyta), filamentous blue-green algae (Cyanobacteria), flagellated green algae (Chlorophyta) and euglenoids (Euglenophyta). Significant levels of PTOX cyanobacteria were observed, including Nodularia sp. (>1000 filaments per mL), Geitlerinema sp. and Anabaena sp.

2 The sample was dominated by flagellated algae, including flagellated green algae (Chlorophyta) and euglenoids (Euglenophyta; ≥3 species). PTOX cyanobacteria observed included Anabaena sp. (≥3 filaments per mL), a cyanophyte filament (per mL) resembling Aphanizomenon sp., and a single oscillatoriacean filament (1 per mL).

Potential toxin producing genera observed include: Microcystins/ Nodularins Saxitoxins  
Anatoxin-a Cylindrospermopsin Anabaena Nodularia Anabaena Geitlerinema Anabaena  
Anabaena

#### Recommendations

Analysis for microcystins/nodularins, anatoxin-a, saxitoxin and cylindrospermopsin is recommended for sample 1. Due to the limited cyanobacteria presence, toxin analysis is currently not recommended for sample 2.

Summary of Results for Analysis from the recommendation above.

Sample ID	MCs/NODs (ng/mL)	CYN (ng/mL)	ANTX-A (ng/mL)	STX (ng/mL)
1 (Lemmon Lake Inlet)	ND	ND	ND	ND
MRL (ng/mL):	0.15	0.05	0.05	0.05
Analyst				
Initials:	KC	MA	MA	KC
Date Analyzed:	8/24/18	8/23/18	8/23/18	8/23/18

ND=Not Detected above MRL

MRL = Method Reporting Limit

Footnotes:

MCs/NODs A microcystins/nodularins Adda ELISA (Abraxis) was utilized for the quantitative and sensitive congener-independent detection of MCs/NODs (US EPA Method 546 & Ohio EPA DES 701.0). The current method reporting limit is 0.15 ng/mL (ppb) based on kit sensitivity, dilution factors and initial demonstration of capability

STX A saxitoxin specific ELISA (Abraxis PN 52255B) was utilized for the detection and quantification of saxitoxin and related analogs (paralytic shellfish toxins – PSTs). The current method reporting limit is 0.05 ng/mL (ppb) based on kit sensitivity and dilution factors. Based on manufacture instructions, the STX ELISA is less cross-reactive to other PSTs and will likely underestimate total PSTs/Saxitoxins. Reported cross-reactivities are as follows: NEO (1.3%), dcSTX (29%), GTX2/3 (23%), GTX5 (23%), dcGTX2/3 (1.4%), dcNEO (0.6%) & GTX1/4 (<0.2%).

ANTX-A (anatoxin-a) Liquid chromatography mass spectrometry/mass spectrometry (LC-MS/MS) ANTX-A The [M+H]<sup>+</sup> ion for ANTX-A (m/z 166) was fragmented and the product ions (m/z 91, 131 & 149) were monitored. The sample response was compared to LFSM response utilizing the quantification ion, m/z 91. An external standard curve was used to determine LFSM returns.

CYN (cylindrospermopsin) The [M+H]<sup>+</sup> ion for CYN (m/z 416) was fragmented and the product ions (m/z 194, 274, 336) were monitored. The sample response was compared to LFSM response utilizing the quantification ion, m/z 336. An external standard curve was used to determine LFSM returns.